



001560-373.ST25

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JUN 11 2002

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SEQUENCE LISTING

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<120> Process for Producing Peptides Using a Helper Peptide

<130> 001560-373

<140> US 09/402,093

<141> 1999-09-29

<150> PCT/JP99/00406

<151> 1999-01-29

<150> JP 10-32272

<151> 1998-01-30

<160> 25

<170> FastSEQ for Windows Version 4.0

<210> 1

<211> 4

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence adjacent to a site cleaved by
enterokinase

<400> 1

Asp Asp Asp Lys

1

<210> 2

<211> 4

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence adjacent to a site cleaved by
blood coagulation Factor Xa

<400> 2

Ile Glu Gly Arg

1

<210> 3

<211> 7

<212> PRT

<213> Artificial Sequence

DI
<220>

<223> Amino acid sequence containing a site cleaved by
renin

<400> 3

Pro Phe His Leu Leu Val Tyr
1 5

<210> 4

<211> 6

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence of helper peptide

<400> 4

Val Asp Asp Asp Lys
1 5

<210> 5

<211> 6

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence of helper peptide

<400> 5

Gly Cys His His His His
1 5

<210> 6

<211> 10

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence containing a chemically
cleaved site

<400> 6

Pro Gly Gly Arg Pro Ser Arg His Lys Arg
1 5 10

<210> 7

<211> 10

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence of helper peptide

<400> 7

His Arg His Lys Arg Ser His His His His
 1 5 10

<210> 8

<211> 5

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence containing a site cleaved by
 Kex2 Protease

<400> 8

Ser Asp His Lys Arg
 1 5

<210> 9

<211> 23

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence containing a position cleaved
 by OmpT

<400> 9

Gln Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His
 1 5 10 15
 Arg Trp Gly Arg Ser Gly Ser
 20

<210> 10

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence containing a position cleaved
 by OmpT

<400> 10

Gln Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His
 1 5 10 15
 Gly Ser Gly Ser
 20

<210> 11

<211> 69

<212> DNA

<213> Artificial Sequence

<220>

<223> Nucleotide sequence coding for an amino acid
sequence containing a site cleaved by OmpT

<221> CDS

<222> (1)...(69)

<400> 11

cag	atg	cat	ggt	tat	gac	gcg	gag	ctc	cgg	ctg	tat	cgc	cgt	cat	cac	48
Gln	Met	His	Gly	Tyr	Asp	Ala	Glu	Leu	Arg	Leu	Tyr	Arg	Arg	His	His	
1				5				10					15			

cgg	tgg	ggt	cgt	tcc	gga	tcc	69
Arg	Trp	Gly	Arg	Ser	Gly	Ser	
			20				

<210> 12

<211> 23

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence containing a site cleaved by
OmpT

<400> 12

Gln	Met	His	Gly	Tyr	Asp	Ala	Glu	Leu	Arg	Leu	Tyr	Arg	Arg	His	His
1				5				10					15		
Arg	Trp	Gly	Arg	Ser	Gly	Ser									
			20												

<210> 13

<211> 47

<212> DNA

<213> Artificial Sequence

<220>

<223> Nucleotide sequence coding for an amino acid
sequence containing a site cleaved by OmpT

<400> 13

tgg	tat	gac	g	c	g	g	a	g	c	t	c	c	g	c	t	g	a	t	a	c	c	g	g	t	t	c	c	g	47
-----	-----	-----	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	----

<210> 14

<211> 55

<212> DNA

<213> Artificial Sequence

<220>

<223> Nucleotide sequence coding for an amino acid
sequence containing a site cleaved by OmpT

<400> 14

gat	c	c	g	a	a	a	c	c	g	t	g	a	t	g	a	c	g	g	a	t	a	c	a	a	c	c	a	t	g	c	a	55
-----	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	---	----

<210> 15

<211> 24
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Primer

<400> 15
 gactcagatc ttcctgagggc cgat 24

<210> 16
 <211> 36
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Primer

<400> 16
 aaaggtacct tccgcatgcc gcggatgtcg agaagg 36

<210> 17
 <211> 20
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Primer

<400> 17
 aggccaggaa ccgtaaaaag 20

<210> 18
 <211> 29
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Primer

<400> 18
 aaaatgcata gcatacgtaac cgtgcatct 29

<210> 19
 <211> 627
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Nucleotide sequence coding for a fusion protein
 comprising GLP-1, helper peptide and
 beta-galactosidase protective peptide

<221> CDS
 <222> (82)...(543)

<400> 19

cccaggcttt	acacttttatg	cttccggctc	gtatgttgtg	tggaattgtg	agcggataac	60			
aatttcacac	aggaaacagc	atg acc	atg att	acg gat	tca ctg	gcc gtc	111		
		Met Thr	Met Ile	Thr Asp	Ser Leu	Ala Val			
		1		5		10			
gtt tta	caa cgt	aaa gac	tgg gat	aac cct	ggc gtt	acc caa	ctt aat	159	
Val Leu	Gln Arg	Lys Asp	Trp Asp	Asn Pro	Gly Val	Thr Gln	Leu Asn		
		15		20			25		
cgc ctt	gca gca	cat ccc	cct ttc	gcc agc	tgg cgt	aat agc	gac gac	207	
Arg Leu	Ala Ala	His Pro	Pro Phe	Ala Ser	Trp Arg	Asn Ser	Asp Asp		
		30		35			40		
gcc cgc	acc gat	cgc cct	tcc caa	cag cag	ttg cgc	agc ctg	aat ggc	gaa	255
Ala Arg	Thr Asp	Arg Pro	Ser Gln	Gln Leu	Arg Ser	Leu Asn	Gly Gly	Glu	
		45		50			55		
tgg cgc	ttt gcc	tgg ttt	ccg gca	cca gaa	gcg gtg	ccg gca	agc ttg		303
Trp Arg	Phe Ala	Trp Phe	Pro Ala	Pro Glu	Ala Val	Pro Ala	Ser Leu		
		60		65			70		
ctg gag	tca gat	ctt cct	gag gcc	gat act	gtc gtc	gtc ccc	tca aac		351
Leu Glu	Ser Asp	Leu Pro	Glu Ala	Asp Thr	Val Val	Val Pro	Ser Asn		
		75		80			85		
tgg cag	atg cac	ggc tac	gat gcg	atg cat	ggc tat	gac gcg	gag ctc		399
Trp Gln	Met His	Gly Tyr	Asp Ala	Met His	Gly Tyr	Asp Ala	Glu Leu		
		95		100			105		
cgc ctg	tat cgc	cgt cat	cac gcc	ggc tcc	gga tcc	cct tct	cga cat	ccg	447
Arg Leu	Tyr Arg	Arg His	His His	Gly Ser	Gly Ser	Pro Ser	Arg His	Pro	
		110		115			120		
cgg cat	gcg gaa	ggc ggt	acc ttt	acc agc	gat gtg	agc tcg	tat ctg	gaa	495
Arg His	Ala Glu	Gly Gly	Thr Phe	Thr Ser	Asp Val	Ser Ser	Tyr Leu	Glu	
		125		130			135		
ggc cag	gcg gca	aaa gaa	ttc atc	gcg tgg	ctg gtg	aaa ggc	cgt ggt		543
Gly Gln	Ala Ala	Lys Glu	Phe Ile	Ala Trp	Leu Val	Lys Gly	Arg Gly		
		140		145			150		
taagtcgaca	gccgcgctaa	tgagcgggct	tttttttctc	ggaattaatt	ctcatgtttg	603			
acagcttatc	atcgataagc	ttta				627			

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<210> 20
<211> 154
<212> PRT
<213> Artificial Sequence
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<220>
<223> Amino acid sequence of a fusion protein comprising
GLP-1, helper peptide and beta-galactosidase
protective peptide

<400> 20
Met Thr Met Ile Thr Asp Ser Leu Ala Val Val Leu Gln Arg Lys Asp
1 5 10 15

Trp Asp Asn Pro Gly Val Thr Gln Leu Asn Arg Leu Ala Ala His Pro
 20 25 30
 Pro Phe Ala Ser Trp Arg Asn Ser Asp Asp Ala Arg Thr Asp Arg Pro
 35 40 45
 Ser Gln Gln Leu Arg Ser Leu Asn Gly Glu Trp Arg Phe Ala Trp Phe
 50 55 60
 Pro Ala Pro Glu Ala Val Pro Ala Ser Leu Leu Glu Ser Asp Leu Pro
 65 70 75 80
 Glu Ala Asp Thr Val Val Val Pro Ser Asn Trp Gln Met His Gly Tyr
 85 90 95
 Asp Ala Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His
 100 105 110
 His Gly Ser Gly Ser Pro Ser Arg His Pro Arg His Ala Glu Gly Thr
 115 120 125
 Phe Thr Ser Asp Val Ser Ser Tyr Leu Glu Gly Gln Ala Ala Lys Glu
 130 135 140
 Phe Ile Ala Trp Leu Val Lys Gly Arg Gly
 145 150

<210> 21
 <211> 187
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> Amino acid sequence of a fusion protein comprising
 GLP-1, helper peptide and beta-galactosidase
 protective peptide

<400> 21
 Met Thr Met Ile Thr Asp Ser Leu Ala Val Val Leu Gln Arg Lys Asp
 1 5 10 15
 Trp Asp Asn Pro Gly Val Thr Gln Leu Asn Arg Leu Ala Ala His Pro
 20 25 30
 Pro Phe Ala Ser Trp Arg Asn Ser Asp Asp Ala Arg Thr Asp Arg Pro
 35 40 45
 Ser Gln Gln Leu Arg Ser Leu Asn Gly Glu Trp Arg Phe Ala Trp Phe
 50 55 60
 Pro Ala Pro Glu Ala Val Pro Ala Ser Leu Leu Glu Ser Asp Leu Pro
 65 70 75 80
 Glu Ala Asp Thr Val Val Val Pro Ser Asn Trp Gln Met His Gly Tyr
 85 90 95
 Asp Ala Pro Ile Tyr Thr Asn Val Thr Tyr Pro Ile Thr Val Asn Pro
 100 105 110
 Pro Phe Val Pro Thr Glu Pro His His His His His Gly Gly Arg Gln
 115 120 125
 Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His Arg
 130 135 140
 Trp Gly Arg Ser Gly Ser Pro Ser Arg His Lys Arg His Ala Glu Gly
 145 150 155 160
 Thr Phe Thr Ser Asp Val Ser Ser Tyr Leu Glu Gly Gln Ala Ala Lys
 165 170 175
 Glu Phe Ile Ala Trp Leu Val Lys Gly Arg Gly
 180 185

<210> 22
 <211> 184
 <212> PRT
 <213> Artificial Sequence

<220>

<223> Amino acid sequence of a fusion protein comprising
 GLP-1, helper peptide and beta-galactosidase
 protective peptide

<400> 22

DI
 Met Thr Met Ile Thr Asp Ser Leu Ala Val Val Leu Gln Arg Lys Asp
 1 5 10 15
 Trp Asp Asn Pro Gly Val Thr Gln Leu Asn Arg Leu Ala Ala His Pro
 20 25 30
 Pro Phe Ala Ser Trp Arg Asn Ser Asp Asp Ala Arg Thr Asp Arg Pro
 35 40 45
 Ser Gln Gln Leu Arg Ser Leu Asn Gly Glu Trp Arg Phe Ala Trp Phe
 50 55 60
 Pro Ala Pro Glu Ala Val Pro Ala Ser Leu Leu Glu Ser Asp Leu Pro
 65 70 75 80
 Glu Ala Asp Thr Val Val Val Pro Ser Asn Trp Gln Met His Gly Tyr
 85 90 95
 Asp Ala Pro Ile Tyr Thr Asn Val Thr Tyr Pro Ile Thr Val Asn Pro
 100 105 110
 Pro Phe Val Pro Thr Glu Pro His His His His His Gly Gly Arg Gln
 115 120 125
 Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His Gly
 130 135 140
 Ser Gly Ser Pro Ser Arg His Lys Arg His Ala Glu Gly Thr Phe Thr
 145 150 155 160
 Ser Asp Val Ser Ser Tyr Leu Glu Gly Gln Ala Ala Lys Glu Phe Ile
 165 170 175
 Ala Trp Leu Val Lys Gly Arg Gly
 180

<210> 23
 <211> 184
 <212> PRT
 <213> Artificial Sequence

<220>

<223> Amino acid sequence of a fusion protein comprising
 GLP-1, helper peptide and beta-galactosidase
 protective peptide

<400> 23

Met Thr Met Ile Thr Asp Ser Leu Ala Val Val Leu Gln Arg Lys Asp
 1 5 10 15
 Trp Asp Asn Pro Gly Val Thr Gln Leu Asn Arg Leu Ala Ala His Pro
 20 25 30
 Pro Phe Ala Ser Trp Arg Asn Ser Asp Asp Ala Arg Thr Asp Arg Pro
 35 40 45
 Ser Gln Gln Leu Arg Ser Leu Asn Gly Glu Trp Arg Phe Ala Trp Phe
 50 55 60
 Pro Ala Pro Glu Ala Val Pro Ala Ser Leu Leu Glu Ser Asp Leu Pro

65					70				75					80	
Glu	Ala	Asp	Thr	Val	Val	Val	Pro	Ser	Asn	Trp	Gln	Met	His	Gly	Tyr
				85					90					95	
Asp	Ala	Pro	Ile	Tyr	Thr	Asn	Val	Thr	Tyr	Pro	Ile	Thr	Val	Asn	Pro
			100					105					110		
Pro	Phe	Val	Pro	Thr	Glu	Pro	His	His	His	His	His	Gly	Gly	Arg	Gln
		115					120					125			
Met	His	Gly	Tyr	Asp	Ala	Glu	Leu	Arg	Leu	Tyr	Arg	Arg	His	His	Gly
	130					135					140				
Ser	Gly	Ser	Pro	Ser	Arg	His	Pro	Arg	His	Ala	Glu	Gly	Thr	Phe	Thr
145					150					155					160
Ser	Asp	Val	Ser	Ser	Tyr	Leu	Glu	Gly	Gln	Ala	Ala	Lys	Glu	Phe	Ile
			165					170						175	
Ala	Trp	Leu	Val	Lys	Gly	Arg	Gly								
			180												

DI
 <210> 24
 <211> 5
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> Amino acid sequence containing a site cleaved by
 Kex2 Protease

<400> 24
 Ser Cys His Lys Arg
 1 5

<210> 25
 <211> 6
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> Amino acid sequence containing a site cleaved by
 Kex2 Protease

<221> PEPTIDE
 <222> 6
 <223> Xaa = Gly

<400> 25
 Arg His His Gly Pro Xaa
 1 5